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⑤④ Methods and materials for treatment of disease states involving Immunological factors.

⑤⑦ In animals including humans, the symptoms of non-naphylactic disease states wherein the disease victim's humoral and/or cell-mediated immunological response is involved in disease pathology are alleviated upon administration of compositions comprising mixtures of histamine and one or more immunogenic substances participative in the humoral or cell-mediated pathogenic response. In preferred embodiments, the compositions are administered in essentially minute, "neutralizing" doses. Illustratively, significant relief of symptoms of multiple sclerosis is achieved through parenteral (e.g., subcutaneous or sublingual) administration of mixtures of histamine and measles virus immunogen. For a majority of patients, from about  $8.8 \times 10^{-6}$  to  $5.5 \times 10^{-3}$  mg of histamine phosphate will provide an effective quantity of histamine to a unit dose of the mixture. Likewise, a unit dose of the mixture will contain from about  $2 \times 10^{-3}$  to about  $4 \times 10^{-4}$  times the prescribed vaccination dose of a killed, attenuated measles virus vaccine strain. As another example, rheumatoid arthritis symptoms are alleviated by administration of histamine admixed with an immunoglobulin G immunogen which is provocative of *in vivo* rheumatoid factor production.

Other immune disorders (e.g., chronic pain and herpes simplex type II infections) are effectively treated with admixtures of histamine and appropriate immunogen(s).

"METHODS AND MATERIALS FOR TREATMENT OF DISEASE  
STATES INVOLVING IMMUNOLOGICAL FACTORS"

BACKGROUND

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The present invention relates generally to the treatment of disease states involving immunological factors and more particularly to methods and materials for alleviation of symptoms of nonanaphylactic disorders wherein disease pathology results in whole or part from the victim's own humoral and/or cell-mediated immune response to one or more immunogenic substances.

10 Disease states involving immunological factors may be seen to broadly comprise (1) immunodeficiency diseases, and (2) disorders wherein tissue injury occurs as a result of a humoral or cell-mediated response to immunogens (e.g., antigens) of endogenous or exogenous origin. This latter group of immunological disorders is frequently referred to as involving immune "hyper-sensitivity", with the numerous disease states comprehended by the term classified according to four hyper-sensitivity "reaction" types.

15 Type I reactions (frequently referred to as anaphylactic, immediate-type, atopic, reagenic, or IgE-mediated hypersensitivity reactions) generally result from the release of pharmacologically active substances such as histamine, slow-reacting substance of anaphylaxis (SRS-A), and eosinophilic chemotactic

factor (ECF) from IgE-sensitized basophils and mast cells after contact with a specific exogenous antigen. Disease states in which Type I reactions play a role include allergic extrinsic asthma, seasonal allergic rhinitis, systemic anaphylaxis, and the like.

5 Type II reactions (also referred to as cytotoxic, cytolytic complement-dependent or cell-stimulating hypersensitivity reactions) result when antibody reacts with antigenic components of cells or tissue elements or with an antigen or hapten which has become  
10 intimately coupled to cells or tissue. Disease states with which Type II reactions are associated include autoimmune hemolytic anemia, erythroblastosis fetalis and Goodpasture's disease.

15 Type III reactions (also referred to as toxic complex, soluble complex, or immune complex hypersensitivity reactions) result from the deposition of soluble circulating antigen-antibody complexes in vessels or in tissues, with accompanying acute  
20 inflammatory reactions at the site of immune complex deposition. Prototypical Type III reaction disorders include Arthus reaction, serum sickness, systemic lupus erythematosus, and certain types of glomerulonephritis.

25 Type IV reactions (frequently called cellular, cell-mediated, delayed, or tuberculin-type hypersensitivity reactions) are caused by sensitized T-lymphocytes which result from contact with a specific antigen. Examples of diseases cited as involving Type IV reactions  
30 are contact dermatitis and allograft rejection.

Of particular interest in consideration of the present invention are those immunological disorders, especially prominent in humans, which are commonly referred to as "autoimmune" diseases. While  
35 the classification originated to distinguish immune disorders involving reaction against "self" or endogenous

antigens from those involving reaction against exogenous antigens, significant controversy exists over the criteria for establishing a particular disease as autoimmune in origin. Autoantibodies can frequently  
5 be demonstrated in a large number of individuals who are free of autoimmune disease and autoimmune diseases such as rheumatic heart disease appear to involve, inter alia, cross-reactive immune responses to antigens of clearly exogenous origins (Streptococcus sp.).  
10 The designation of a particular disease as an autoimmune disorder is often subjectively based on (1) presence of some evidence of an antigen-antibody reaction including an endogenous antigenic substance, (2) judgement that the immunologic findings are not merely secondary  
15 to some other disease state, and (3) the lack of other identifiable causes for the disorder.

While autoimmune disorders are frequently associated with Type II hypersensitivity reactions, there exists evidence of involvement of Type III and  
20 Type IV reactions in some autoimmune diseases. Essentially as a matter of definition, immunological disorders of the "anaphylactic" or IgE-mediated type (i.e., Type I hypersensitivity reactions) are not classified as autoimmune due to the clear involvement  
25 of clearly exogenous antigenic substances.

Autoimmune diseases are frequently characterized by means of their involvement of single organ or single cell-types or involvement of multiple organs or tissue systems. (Due to the frequent involvement  
30 of either connective tissue and blood vessels in the diseases of the latter, systemic, type, these have frequently been referred to as "collagen," or "collagen-vascular" or "connective tissue" diseases.)

Diseases frequently designated as single  
35 organ or single cell-type autoimmune disorders include: Hashimoto's thyroiditis, autoimmune hemolytic anemia,

autoimmune atrophic gastritis of pernicious anemia,  
autoimmune encephalomyelitis, autoimmune orchitis,  
Goodpasture's disease, autoimmune thrombocytopenia,  
sympathetic ophthalmia, myasthenia gravis, Graves'  
5 disease, primary biliary cirrhosis, chronic aggressive  
hepatitis, ulcerative colitis and membranous glomeru-  
lopathy. Diseases often designated as involving systemic  
autoimmune disorder include: systemic lupus erythematosus,  
rheumatoid arthritis, Sjögren's syndrome, Reiter's  
10 syndrome, polymyositis-dermatomyositis, systemic sclerosis,  
polyarteritis nodosa and multiple sclerosis.

It is unfortunately the case that the treat-  
ment of many of the above-noted autoimmune disorders  
is, at best, primitive and seldom effective in alleviat-  
15 ing disease symptoms for significant periods of time.  
Progressive degenerative autoimmune diseases such  
as systemic lupus erythematosus, systemic sclerosis,  
polyarteritis, polymyositis and multiple sclerosis  
are usually treated by administration of high doses  
20 of corticosteroids to alleviate discomfort due to  
inflammation of tissues. Immunosuppressant therapy  
has proven to be of little value. Drug therapy (see,  
e.g., U.S. Patent Nos. 3,864,481 and 4,113,858) has  
provided no consistent alleviation of the frequently  
25 painful symptoms nor altered substantially the general  
prognosis of progressive debilitation and death.

The following somewhat disparate items of  
information concerning the state of the art in immunology  
are of considerable interest to the background of  
30 the invention.

Histamine [4-(2-aminoethyl)-imidazole or  
 $\beta$ -imidazolylethylamine] is a chemical substance possess-  
ing pronounced biological activities. It is a powerful  
stimulant of gastric secretion and constrictor of  
35 smooth muscle. It is a vasodilator and large doses  
cause a relatively rapid fall in blood pressure.

Paradoxically, however, it has been shown that histamine

It is frequently noted to be liberated by epithelial cells upon traumatic injury or stimulation by antigenic substances. Its storage in and release from mast cells and basophil granules and its role in Type I hypersensitivity reactions have been the subject of extensive study. At present, its clinical use is principally as a stimulant in diagnosis of impairment of gastric secretion. From time to time, therapeutic uses have been proposed including administration for alleviation of certain kinds of headache symptoms. With the single exception of an experimental study into use of homeopathic doses in treatment of multiple sclerosis [Jones, "My Fight to Conquer Multiple Sclerosis," Messner (1952)] it has not been employed in the treatment of any autoimmune disorders.

To the extent that autoimmune diseases have been the subject of etiological investigation, it has frequently been the case that, with varying degrees of precision, one or more endogenous or exogenous immunogens has been associated with participation in formation of circulating antibodies. See, e.g., Holborow, et al., "Immunoassays in Autoimmune Disease" appearing at pp. 277-288 in "Immunoassays for the 80's," Voller, et al., eds., University Park Press, Baltimore (1981). Results of such studies have been principally of diagnostic significance and have been of little use in formulating therapeutic regimens for the diseased patients. As one example, there have been numerous reports of isolation of circulating (humoral) antiviral antibodies from multiple sclerosis patients, including antibodies against vaccinia, rubella, herpes simplex, varicella-zoster, mumps, Epstein-Barr virus, adenovirus and even canine distemper virus. Despite the lack of any evidence of viral particle involvement in lesions or cerebrospinal fluids of multiple sclerosis patients, one of the most consistently

reported antiviral antibodies found in such patients has been antimeasles virus antibody. Further, the applicant and his co-workers were able to employ homogenized lymphocytes of multiple sclerosis patients to  
5 provoke formation of heterologous species antibodies which, in turn, have been found to be exceptionally useful in the development of diagnostic procedures. See, e.g., U.S. Letters Patent No. 4,294,818. This lymphocyte-associated immunogen, (i.e., the antigen  
10 responsible for formation of the highly specific antibodies reactive with sera of all multiple sclerosis patients) has not yet been purified to homogeneity and characterized, nor has it been employed in any form in any known therapeutic regimen for alleviation  
15 of multiple sclerosis symptoms. As another example, the rheumatoid arthritis disease state is frequently proposed as having an autoimmune basis on the ground of a characteristic presence of circulating "RF" (rheumatoid factor), an anti-immunoglobulin antibody directed  
20 at IgG. In seropositive patients, it is proposed that RF, which can undergo a complement fixing interaction with IgG, is involved in inducing and maintaining chronic synovitis.

Finally, over the last twenty or so years,  
25 a small body of information has developed concerning a therapeutic regimen known as "provocative neutralization" therapy. See, e.g., Miller, "Food Allergy, Provocative Testing and Injection Therapy," Charles C. Thomas, Springfield, Illinois (1972); Miller, Annals of Allergy, 38, pp. 185-191 (1977); Miller, Trans.Am.Soc. Opth.&Otolar.Allergy, 14, pp. 159-168 (1974); and  
30 Miller, Clinical Med., 81, pp. 16-19 (1974). Briefly put, the therapeutic scheme involves subcutaneous administration of a known "causative" agent of a particular disease state in amounts comprising the patient's  
35 maximum tolerated intradermal dose. This dose is

defined as the smallest dilution (i.e., highest concentration) of the substance which will produce a negative wheal when employed in a standard skin test injection procedure. The proposed mode of operation of this regimen is through modulation of sub-populations of T-lymphocytes. Initial successes for this treatment procedure were reported in the context of relief of headache symptoms correlated with food allergies. Subsequently, relief of symptoms of acute influenza infection has been reported to attend provocative neutralization treatments with live attenuated influenza virus. Further, relief from herpes simplex Type I infection has been reported for patients treated with influenza virus.

15

#### BRIEF SUMMARY

Provided by the present invention are novel methods and materials for obtaining significant alleviation of symptoms of nonanaphylactic disease states wherein tissue injury results from the disease victim's own humoral and/or cell-mediated immunological response to one or more immunogenic substances of endogenous or exogenous origin. Compositions administered according to the invention comprise mixtures of histamine and one or more of the immunogenic substances specifically immunologically associated with the disease state (as indicated by, e.g., characteristic elevation of circulating antibodies thereto in the disease victim). The compositions are administered in small, "neutralizing" doses. The mixtures may be provided in any immunologically acceptable diluent, with or without immunological adjuvants.

As examples of the practice of the invention, a study of the effects of administration of a histamine phosphate and measles virus mixture to a large number



of multiple sclerosis patients has revealed remarkably high percentages of patients reporting substantial improvement in disease symptoms. Rheumatoid arthritis patients treated according to the invention with compositions including histamine and, for example, rabbit anti-sheep red blood cell immunoglobulin G (IgG) which is provocative of rheumatoid factor (RF) formation in humans have uniformly reported relief of arthritis symptoms.

10           According to another aspect of the invention, relief of disease state symptoms has been achieved with respect to disorders not classically described as directly involving immunological factors. As an example, patients experiencing chronic pain of a dura-  
15   tion of three months or more (having a history of either osteoarthritis or major physical trauma) have all reported relief of pain upon treatment with mixtures of histamine and inactivated, attenuated measles virus. In a like manner, patients experiencing recurrent  
20   herpes simplex virus type II infection have reported relief of lesion pain and lesion enlargement upon treatment with compositions including histamine, measles inactivated, attenuated virus and influenza vaccine (killed) virus.

25           The histamine component of mixtures prepared according to the present invention is suitably provided in the form of a water soluble histamine salt. The presently preferred form of histamine is histamine phosphate. For a majority of patients, from about  
30    $8.8 \times 10^{-6}$  to  $5.5 \times 10^{-3}$  mg and preferably  $4.4 \times 10^{-5}$  to  $1.1 \times 10^{-3}$  mg of histamine phosphate will be effective.

          Expectedly, the immunogen components of mixtures may be provided in forms of varying purity  
35   or homogeneity. As one example, measles virus immunogen may be provided to mixtures of the invention in the

form of a inactivated, attenuated measles virus vaccine or as an immunologically active measles virus fraction (e.g., surface antigen preparation) derived from such inactivated, attenuated vaccine strains of virus.

5 Mixtures including histamine and from about  $2 \times 10^{-3}$  to about  $4 \times 10^{-4}$  times the prescribed vaccination dose of killed, attenuated measles virus have been found to be quite useful. In a like manner, the mixture employed in treatment of rheumatoid arthritis may  
10 include Immunoglobulin G as described above or, e.g., the so-called "antigenic," FC region of the heavy IgG chain or a synthetic replica thereof. Mixtures including from 0.1 to 0.005 mg of chromatographically pure IgG have proven to be useful.

15 Positive therapeutic results to date have attended the administration of histamine and immunogen as mixture components, but have not been noted when these substances were individually administered, indicating a type of synergistic, joint activity or formation  
20 of a histamine/immunogen complex which can act as a regulatory molecule for gene expression. This is consistent with the regulatory activity of other well-defined low molecular weight substances.

One route of parenteral administration practiced according to the invention is the subcutaneous  
25 injection of the compositions. Alternately, the compositions may be administered sublingually with good results. Pharmaceutically acceptable diluents, adjuvants and carriers may suitably be employed.

30 Other aspects and advantages of the present invention will be apparent upon consideration of the following detailed description of illustrative examples of the practice thereof.

DETAILED DESCRIPTION

According to the present invention novel compositions are provided which comprise mixtures  
5 of histamine and one or more immunogens. These compositions, in turn, are useful as therapeutic agents in the alleviation of the symptoms of disorders wherein the disease victim's own humoral and/or cell-mediated immunological response is involved in the disease  
10 pathology.

Histamine employed in the present invention is preferably provided in the form of a water soluble histamine salt such as histamine phosphate, salicylate, dihydrochloride, or the like. A suitable histamine  
15 source for use in formulating mixtures of the invention is available from Eli Lilly as "Histamine Phosphate Injection, U.S.P." which is provided in 5 ml units, each ml containing 0.55 mg of histamine phosphate (equivalent to 0.2 mg histamine base) with 16 mg glycerin  
20 and 2 mg phenol.

As employed herein with respect to mixture components of the invention, "immunogen" shall be defined as a substance which, upon introduction into an animal, is capable of stimulating an immune response.  
25 As such, the term includes, but is not limited to, "antigens" which combine with antibodies and/or provoke formation of antibodies in the animal.

One or more immunogens may be provided to mixtures of the invention in forms of widely varying  
30 purity or homogeneity. Where the immunogen employed is of viral origin, for example, it may be provided in the form of whole, inactivated virus particles. Expectedly, much preferred forms of live viruses are attenuated strains commonly employed as non-disease-  
35 producing but nonetheless immunogenic vaccine components. Further, wherever possible, isolated immunogenic frag-

ments of viruses or viral fragment replicas (such as surface antigen isolates or replicas thereof) would be most preferred unless it is established that viral RNA or DNA provides the principal immunogen of interest.

5 As another example, where the immunogen of interest is an antigen which is a constituent of an antibody substance (e.g., an antigenic portion of an immunoglobulin) it may be provided as a pure antigenic isolate from the antibody, or a synthetic replica of such  
10 an antigen, or in the form of the "whole" antibody.

Selection of one or more immunogens for incorporation into a mixture with histamine is made in the context of "specific immunological association" with the particular disorder treated. A principal  
15 factor in the selection process can be the independent determination of the existence of an established correlation between clinical symptoms of the disease state and increased levels of circulating antibodies to the substance in the victim. Another selection factor  
20 is the specific association of an immunogen with the disease state independently of the notation of high levels of circulating antibodies thereto. The multiple sclerosis (MS) disease and the treatment thereof according to the invention provides an illustration of application of such selection criteria in determining immunogen  
25 components. As previously noted, MS patients inexplicably display a characteristic high level of circulating anti-measles antibodies. MS patients also uniformly possess a lymphocyte-associated antigen which is reactive  
30 with certain specific antibodies as described in the diagnostic procedures of U.S. Patent No. 4,294,818. Both measles virus and the lymphocyte-associated antigen therefor qualify as immunogens "specifically immunologically associated" with the MS disease state which  
35 are suitable for combination with histamine in formulation of compositions of the invention useful in alleviating MS disease state symptoms.

The following is a listing of disease states treatable according to the invention and immunogens specifically immunologically associated with the disease state.

5

	<u>Disease State</u>	<u>Immunogen(s)</u>
	1. Rheumatoid arthritis	Immunoglobulin G provocative of RF formation
10	2. Autoimmune hemolytic anemia	Red blood cell surface antigen
	3. Glomerulonephritis associated with $\beta$ hemolytic streptococcal infection	Streptococcal cell surface antigen
15	4. Myasthenia gravis	Acetylcholine receptor
	5. Systemic lupus erythematosus	Human DNA, human epidermal cells, measles virus RNA
	6. Pernicious anemia	Intrinsic factor, gastric mucosal cells
20	7. Hashimoto's thyroiditis	Thyroglobulin
	8. Systemic sclerosis (scleroderma)	Human DNA, immunoglobulin G provocative of RF formation
	9. Polymyositis	Human myoglobin
	10. Sjögren's Syndrome	Thyroglobulin
25	11. Juvenile diabetes	Beta cells

Mixtures of the invention are generally administered parenterally (e.g., subcutaneously intramuscularly, sublingually, etc.) although oral administration is contemplated when provision is made to protect the active ingredients from digestive degradation. Subcutaneous administration is preferred, as is sublingual administration. For example, a typical unit dosage form for use in subcutaneous administration would comprise a mixture of a total volume of 0.5 cc of an immunologically acceptable carrier fluid (prefer-

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35

ably normal saline) to which has been added a histamine source (preferably as a histamine phosphate solution as previously described) and one or more immunogens (e.g., an inactivated, attenuated measles virus vaccine).

- 5 A unit dose for sublingual administration would provide the same quantity of histamine-and-immunogen-mixture in a single drop (typically 0.05 cc) form.

It is contemplated that varying strengths of the histamine component of the pharmaceutical (biologic) mixtures may be formulated. The attending physician may prescribe, or administer, different levels of histamine reflecting alterations in disease states. This is consistent with the procedure of administration of a "neutralizing" dose which is that concentration which specifically immunosuppresses that part of the patient's prevailing immunological response directed against the agent.

- Substantial experience in practice of the invention has revealed that an effective unit dose quantity of histamine for adult patients will be an essentially minute quantity within the range of from about  $8.8 \times 10^{-6}$  to about  $5.5 \times 10^{-3}$  mg and usually between  $4.4 \times 10^{-5}$  and  $1.1 \times 10^{-3}$  mg. The quantity of immunogen employed will depend upon the relative purity or homogeneity of the immunogen. As one example, treatment of MS patients according to the present invention has involved use of unit doses containing from  $2 \times 10^{-3}$  to  $4 \times 10^{-4}$  times the prescribed vaccination dose of attenuated measles virus, vaccine which is generally set at 1000 TCID<sub>50</sub>. Further, a substantial basis exists for believing that the significant, effective immunogenic viral component is a fraction of the virus whose stability is not dependent on the virus being alive at the time of administration.
- 35 [Indeed, in order to eliminate the potential complicating effects of active measles infection, the virus

vaccine preparation employed was allowed to remain at room temperature in phenolated saline over a time of about 3 hours, generally sufficient to kill live virus particles.] Future isolation of the immunologically active, immunogenic fraction(s) of the virus will allow for administration of even smaller weights of substances containing essentially the same quantities of immunogen or synthetic replicas thereof.

As previously noted, administration of histamine and immunogen at separate sites without prior admixture of the components has not proven to have the same effectiveness as single-site administration of mixtures of the components. Thus, while it is possible that beneficial effects may attend parenteral (e.g., subcutaneous) administration of histamine followed by administration of immunogen at the same site (to form a mixture in the patient's skin), such practices are not preferred. The value of admixture of components prior to administration has also been substantiated by the results attending sublingual administration. In treatment of MS patients with histamine and measles virus, it has been generally noted that sublingual administration of a given dose is not as effective as the same subcutaneous dose if the mixture administered is only freshly prepared. Effectiveness of sublingual administration is enhanced by the mere expedient of allowing the mixture of components to stand for a period of time prior to administration.

While not intended to be binding with respect to practice of the invention, a hypothesis exists which would explain the basis for these observations. That the administration of the histamine/immunogen mixture is more effective than separate administration of individual components may be the result of the components' forming a highly active complex of two individually less active components. The effectiveness

of "fresh" mixtures in subcutaneous administration routes may be the result of an almost immediate complex association of histamine with the immunogen. The observation of increased sublingual effectiveness for mixtures which have stood for extended periods of time (vis-a-vis "fresh" mixtures) may be the result of initial formation of a relatively high molecular weight active complex which is not readily absorbed sublingually, with prolonged standing providing somewhat degraded, lower molecular weight, more easily absorbed, but nonetheless active complex.

The following illustrative example relates to practice of the invention in the alleviation of symptoms of multiple sclerosis patients. In the course of the studies set out in the example, it was determined that all multiple sclerosis patients benefitted from administration of admixtures of histamine and immunogen which were within the dose ranges set out above, thereby eliminating the need for individual sensitivity testing carried out in the initial stages of the therapeutic program.

#### EXAMPLE 1

One hundred and sixty-two patients clinically diagnosed as suffering from multiple sclerosis were treated in a therapeutic program of the invention.

The first 24 of the patients were initiated in a program involving administration of a composition which included three mixture components: (1) histamine phosphate (USP Injection, Eli Lilly); (2) measles virus vaccine (Live, Attenuated, MSD, Attenuvax®, Merck, Sharp & Dohme); and influenza virus vaccine (Trivalent Types A and B, Wyeth). Employing a progressive series of five-fold dilutions of the components as received from the manufacturers, each patient was



5 skin tested to determine the maximum immunologically  
tolerated intradermal dose of each component. In  
the case of the measles virus, the commercial vaccine  
was permitted to stand in phenolated saline at room  
temperature for 3 hours, an amount of time believed  
to be sufficient to prompt inactivation of all living  
virus particles. The three mixture components were  
then "custom mixed" for each patient to allow for  
the provision of the indicated dose of each component  
10 in the form of 0.5 cc of fluid. Sublingual administra-  
tion was accommodated by formulation providing the  
indicated dose in only 0.05 cc of fluid (one drop).

15 Patients were periodically required to report  
improvement, worsening or the absence of change with  
respect to twenty motor, neurologic and psychological  
difficulties commonly associated with the multiple  
sclerosis disease state. No report was required if  
the enumerated difficulty had not been encountered  
prior to treatment. Patients were also asked to indi-  
20 cate overall improvement, worsening or absence of  
change with respect to the totality of their disease  
symptoms.

After about two months of treatment in the  
program, certain of the patients were switched to  
25 treatment with two-component mixtures which included  
histamine and either measles virus or influenza virus,  
but not both. Those receiving the measles/histamine  
combination continued to show improvements on par  
with those seen with the three-component mixtures,  
30 while those receiving the histamine/influenza virus  
mixture did not. Consequently, except as indicated  
hereafter, the 24 initial patients and all subsequent  
multiple sclerosis patients in the program received  
the two-component, histamine/measles virus, composition.

35 Certain of the patients were given histamine  
and measles virus in the form of separate subcutaneous

injections at different sites. Upon notation of cessation of alleviation of disease symptoms, these patients were restored to administration of the two components in mixed form.

5                Certain patients were given two-component mixtures which did not include both histamine and measles. All such mixtures failed to generate improvements in disease symptoms on par with those observed for the combination of measles and histamine and all  
10 patients so tested were restored to the measles/histamine combination. The two-component mixtures tested included histamine and either attenuated mumps virus or crude human leukocyte interferon or myelin basic protein (Eli Lilly). Similarly, mixtures of measles  
15 virus vaccine and either of two synthetic precursors of histamine (imidazole and imidazole acetate) were not as effective in relieving disease symptoms.

              Patients were added to the study over the course of nearly eighteen months, until a total of  
20 166 had been enrolled and received at least some treatments. As of January, 1982, 133 patients remained in the study. Of a total of 29 patients who dropped out of the study, two left before completing two months of treatment or providing any evaluation of treatment  
25 effects. Two died of causes unrelated to their multiple sclerosis disease state. Of the twenty-five patients electing to leave the study, only one had reported an overall worsening of symptoms and the remaining  
24 reported either no change (16) or an improvement  
30 (8).

              Table I below tabulates the reports provided by the remaining 133 patients just prior to January, 1982.

TABLE I

Symptom (Number to Which Applicable)	Length of Time with MS Before Treatment (yrs)	Worse (%)	No Change (%)	Better (%)
1. Strength and Endurance (130)	0-5 5-10 10+	9.4 14.3 7.9	28.1 28.6 28.6	62.5 57.1 63.5
2. Energy (132)	0-5 5-10 10+	9.1 13.9 9.5	24.2 25.8 21.7	66.7 58.3 58.7
3. Headache ( 47)	0-5 5-10 10+	0.0 23.1 9.1	58.3 23.1 36.4	41.7 53.8 54.5
4. Double Vision ( 47)	0-5 5-10 10+	7.1 5.6 0.0	35.7 33.3 50.0	57.1 61.1 50.0
5. Blurred Vision ( 64)	0-5 5-10 10+	5.0 9.1 0.0	55.0 40.9 45.5	40.0 50.0 54.5
6. Dizziness ( 56)	0-5 5-10 10+	10.5 11.8 10.0	52.6 35.3 35.0	36.8 52.9 55.0

TABLE I (cont'd.)

Symptom (Number to Which Applicable)	Length of Time with MS Before Treatment (yrs)	Worse (%)	No Change (%)	Better (%)
7. Difficulty Swallowing: Choking ( 62)	0-5 5-10 10+	20.0 5.6 0.0	20.0 44.4 53.6	60.0 50.0 46.4
8. Constipation ( 85)	0-5 5-10 10+	0.0 12.0 2.7	52.2 48.0 40.5	47.8 40.0 56.8
9. Bowel Urgency (control) ( 81)	0-5 5-10 10+	5.0 12.5 0.0	40.0 45.8 48.6	55.0 41.7 51.4
10. Bladder Frequency (113)	0-5 5-10 10+	7.1 9.7 5.6	39.3 38.7 14.8	53.6 51.6 79.6
11. Bladder Urgency (control) (110)	0-5 5-10 10+	7.4 9.7 3.8	37.0 38.7 32.7	55.6 51.6 63.5
12. Sexual Interest and Function ( 74)	0-5 5-10 10+	16.7 18.2 2.9	66.7 63.6 79.4	16.7 18.2 17.6

TABLE I (cont'd.)

Symptom (Number to Which Applicable)	Length of Time with MS Before Treatment (yrs)	Worse (%)	No Change (%)	Better (%)
13. Finger Dexterity; Hand Coordination (110)	0-5 5-10 10+	11.5 12.9 7.5	61.5 48.4 43.4	26.9 38.7 49.1
14. Pain - Any Location ( 64)	0-5 5-10 10+	16.7 31.3 10.0	44.4 50.0 50.0	38.9 18.8 40.0
15. Numbness - Any Location (102)	0-5 5-10 10+	10.0 8.0 8.5	46.7 56.0 57.4	43.3 36.0 34.0
16. Depression ( 77)	0-5 5-10 10+	9.5 23.1 3.3	52.4 34.6 33.3	38.1 42.3 63.3
17. Balance (123)	0-5 5-10 10+	15.6 29.4 10.5	46.9 41.2 52.6	37.5 29.4 36.8
18. Walking (109)	0-5 5-10 10+	23.1 19.4 13.5	50.0 58.1 55.8	26.9 22.6 [30.6]

TABLE I (cont'd.)

Symptom (Number to Which Applicable)	Length of Time with MS Before Treatment (yrs)	Worse (%)	No Change (%)	Better (%)
19. Speech ( 71).	0-5 5-10 10+	17.6 18.2 6.5	41.2 40.9 41.9	[41.2] [40.9] [51.6]
20. Spasms or Tremors (101)	0-5 5-10 10+	20.0 16.7 15.7	20.0 26.7 39.2	60.0 56.7 45.1
21. Overall Response 128 *	0-5 5-10 10+	9.1 5.9 4.9	18.2 17.6 21.3	[72.7] [76.5] [73.8]

\* 5 patients did not respond.

Table II below provides an analysis of the number of unit doses administered in the course of therapy. All patients were initiated in a subcutaneous injection program. Some were changed to either drops  
5 alone or a combination of shots and drops. A total of 45 of the 111 patients who tried the sublingual drops at one time in the program did not return to the subcutaneous injection regimen.

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TABLE II

Route of Administration	Total Patients	Less Than 1 per 2 days (%)	1 per 2 days (%)	1 Daily (%)	2 Daily (%)	3 Daily (%)	More Than 3 Daily (%)
Injection	133	4.5	11.2	53.7	26.8	3.7	0.0
Drop (only)	47	0.0	2.1	17.0	48.9	23.4	8.5
Mixed Shot/Drop	23	0.0	0.0	48.0	52.0	0.0	0.0



- 24 -

The following example illustrates practice of the invention in treatment of rheumatoid arthritis disease symptoms. As in the previous example, it was determined that for all patients the neutralizing levels of the histamine admixture component fell within the concentration levels previously described.

EXAMPLE 2

Six patients clinically diagnosed as suffering from rheumatoid arthritis were initiated in a therapeutic program according to the invention. Each patient was tested to determine the maximum immunologically tolerated intradermal dose of (1) histamine phosphate (USP Injection, Eli Lilly); (2) measles virus vaccine (Live, Attenuated, MSD, Attenuvax\*, Merck, Sharp & Dohme); and (3) rheumatoid factor-provoking, rabbit anti-sheep red blood cell IgG (Cappel Labs). Based on test results, compositions were formulated by admixture of the three components so as to provide the indicated amount of each component in either a 0.5 cc subcutaneously injectable fluid form or a 0.05 cc fluid droplet form for sublingual administration. The frequency of administration varied from one injection every two days to two injections per day, or 1 to 2 sublingual droplet administrations per day. After 6 months of treatment, all six patients have reported improvement in terms of: (1) decreased pain; (2) increased mobility and flexibility; (3) decreased swelling; (4) decreased inflammation; and (5) increased energy. Certain of the patients were subsequently changed over to a regimen wherein a histamine/IgG mixture (without measles virus) was administered and all these continued to show improvement, indicating that the measles virus component is not essential for rheumatoid arthritis treatment.

Favorable responses were noted for use of admixtures including from 0.1 to 0.005 mg of chromatographically pure IgG was employed. Preliminary data indicates that human IgG is as effective as the above-

Examples 1 and 2 are believed to illustrate the applicability of the present invention to treatment of a variety of disease states other than multiple sclerosis and rheumatoid arthritis. Indeed each of the ten additional specific states of an "autoimmune" character previously enumerated as well as other immune disorders are believed to be treatable with similar good results by administration of effective amounts of admixture compositions consisting of histamine and one or more listed immunogens which are specifically immunologically associated with the disease state.

The following example relates to practice of the invention in alleviation of the symptoms of disease states which are not classically described as involving an immune component.

### EXAMPLE 3

Fifteen human patients experiencing chronic pain (i.e., pain without substantial remission over a period of three months or more) were initiated in a therapeutic program. Five of the patients had previously been diagnosed as having osteoarthritis and the remaining ten had initially experienced pain subsequent to severe physical trauma. Each patient was tested to determine the maximum tolerated intradermal dose of histamine phosphate and inactivated attenuated measles virus vaccine as in the preceding examples. In each instance, the histamine admixture component concentration determined was in the range of from  $8.8 \times 10^{-6}$  to  $5.5 \times 10^{-3}$  mg of histamine phosphate

per dose and the measles admixture component dose concentration was in the range of  $2 \times 10^{-3}$  to  $4 \times 10^{-4}$  times the prescribed vaccination dose of virus. The frequency of administration varied from one injection  
5 every 4 days to two injections per day, and/or 1 to 3 sublingual drops per day. All patients experienced remission of pain, with the onset of remission occurring in some cases as soon as a few hours after the first administration of the composition.

10 Two human patients suffering from recurrent herpes simplex virus type II infections were skin tested to determine maximum tolerated intradermal doses of histamine phosphate, measles virus vaccine and influenza virus vaccine in the manner of the pre-  
15 vious examples. Unit dose concentrations for histamine phosphate and for each of the virus preparations were within the ranges described above. Both patients received subcutaneous injections of the three-component mixture compositions on a daily basis and reported  
20 immediate remission of lesion pain, followed by decrease in size and eventual elimination of lesions within 2 to 3 days.

A total of ten horses exhibiting moderate to severe lameness were skin tested with histamine  
25 phosphate and measles virus vaccine. Each was then treated with a mixture of the indicated quantities which were in the ranges previously noted. Rapid recovery was observed in all animals and all have remained symptom-free upon continuation of treatment  
30 on regimens varying from 1 injection per day to 1 injection per week.

Numerous modifications and variations of the invention as described in the above illustrative examples are expected to occur to those skilled in  
35 the art. Consequently, only such limitations as appear in the appended claims should be placed thereon.

CLAIMS

1. A composition for use in alleviating the symptoms of  
5 a non-anaphylactic disease state wherein the disease  
victim's humoral and/or cell mediated immunological res-  
ponse is participative in the disease pathology, which  
composition comprises a mixture of histamine and one or  
10 more immunogens specifically immunologically associated  
with the disease state.
2. A composition according to Claim 1, wherein the  
disease state to be treated is multiple sclerosis and an  
immunogen in the composition is inactivated, attenuated  
15 measles virus or an immunologically active fraction  
thereof, and/or an immunologically active constituent of  
homogenized lymphocytes of multiple sclerosis disease  
patient(s).
- 20 3. A composition according to Claim 1, wherein the  
disease state to be treated is rheumatoid arthritis and  
an immunogen in the composition is immunoglobulin G  
provocative of RF formation or an immunologically active  
fraction thereof.
- 25- 4. A composition according to Claim 3, which comprises,  
in unit dosage form, from 0.1 to 0.005 mg of chromatogra-  
phically pure immunoglobulin G.
- 30 5. A composition according to Claim 4, wherein the  
immunoglobulin G is rabbit anti-sheep red blood cell  
immunoglobulin G.
- 35 6. A composition according to Claim 1, wherein the  
disease state to be treated is autoimmune hemolytic  
anemia and an immunogen in the composition is red blood  
cell surface antigen or an immunologically active frac-

tion thereof.

7. A composition according to Claim 1, wherein the disease state to be treated is glomerulonephritis associated with  $\beta$ -hemolytic streptococcal infection and an immunogen in the composition is a streptococcal cell surface antigen or an immunologically active fraction thereof.
- 10 8. A composition according to Claim 1, wherein the disease state to be treated is myasthenia gravis and an immunogen in the composition is acetylcholine receptor or an immunologically active fraction thereof.
- 15 9. A composition according to Claim 1, wherein the disease state to be treated is systemic lupus erythematosus and an immunogen in the composition is human DNA or human epidermal cells or measles virus RNA or an immunologically active fraction thereof.
- 20 10. A composition according to Claim 1, wherein the disease state to be treated is pernicious anemia and an immunogen in the composition is intrinsic factor or gastric mucosal cells or an immunologically active
- 25 fraction thereof.
11. A composition according to Claim 1, wherein the disease state to be treated is Hashimoto's thyroiditis and an immunogen in the composition is thyroglobulin or an immunologically active fraction thereof.
- 20 12. A composition according to Claim 1, wherein the disease state to be treated is systemic sclerosis and an immunogen in the composition is human DNA or immunoglobulin G provocative of RF or an immunologically
- 35 active fraction thereof.

13. A composition according to Claim 1, wherein the disease state to be treated is polymyositis and an immunogen in the composition is human myoglobin or an immunologically active fraction thereof.

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14. A composition according to Claim 1, wherein the disease state to be treated is Sjögren's Syndrome and an immunogen in the composition is thyroglobulin or an immunologically active fraction thereof.

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15. A composition according to Claim 1, wherein the disease state to be treated is juvenile diabetes and an immunogen in the composition is  $\beta$  cells or an immunologically active fraction thereof.

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16. A composition for use in alleviating chronic pain symptoms, which composition comprises a mixture of histamine and inactivated, attenuated measles virus or an immunologically active fraction thereof.

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17. A composition according to Claim 2 or 15, wherein the measles virus is an inactivated, attenuated vaccine strain.

25 18. A composition according to Claim 17, which comprises, in unit dosage form, from about  $2 \times 10^{-3}$  to about  $4 \times 10^4$  times the prescribed vaccination dose of an inactivated, attenuated measles virus vaccine strain.

30 19. A composition according to any one of the preceding claims, wherein histamine is in the form of a water soluble histamine salt.

35 20. A composition according to Claim 19, which comprises, in a unit dosage form, from  $8.8 \times 10^{-6}$  to  $5.5 \times 10^{-3}$  mg of histamine phosphate.

21. A composition for use in alleviating the symptoms of a herpes simplex virus type II disease state, which composition comprises a mixture of histamine, attenuated, inactivated measles virus or an immunologically active fraction thereof, and inactivated influenza virus or an immunologically active fraction thereof.

22. A composition according to Claim 21, wherein histamine is in the form of a water soluble histamine salt and wherein both the measles virus and influenza virus are vaccine strains.

23. A composition according to Claim 22, which comprises, in unit dosage form, from  $8.8 \times 10^{-6}$  to  $5.5 \times 10^{-3}$  mg of histamine phosphate and from  $2 \times 10^{-3}$  to  $4 \times 10^{-4}$  times the prescribed vaccination dose of each of the vaccine strains.





## ANHANG

## ANNEX

## ANNEXE

zum internationalen Recherchen-  
bericht über die internationale  
Patentanmeldung Nr.

to the International Search  
Report to the International Patent  
Application No.

au rapport de recherche inter-  
national relatif à la demande de brevet  
international n°

PCT/US 94/08825 SAE 95995

In diesem Anhang sind die Mitglieder  
der Patentfamilien der im obenge-  
nannten internationalen Recherchenbericht  
angeführten Patentdokumente angegeben.  
Diese Angaben dienen nur zur Unter-  
richtung und erfolgen ohne Gewähr.

This Annex lists the patent family  
members relating to the patent documents  
cited in the above-mentioned inter-  
national search report. The Office is  
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La présente annexe indique les  
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